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Q T V C L51

KPKKIVNAKK D<sub>16</sub> V<sub>17</sub>V N

Tripple alpha helical coiled coil

Exon 2

forming

Trimer stabilising

Exon 1

(B)

ACGUCTCCTGAAGGGATCCTAA Amp - AGCTTGAATTC Hind III Eco R Hind II GATCAATCCAGGGAAGATCTCCTGGTACCGAGCCACCAACC pT7H6 G S I Q G R S P G T E P T T GATCAATCCAGGGAAGATCTCCTGGTACCGAGCCACCAACC Bam HI/Bcl I Bam HI CATATGGGATCGCATCACCATCACCATCACG ρı 回 EH Kon = H Ö Ħ promoter Phage T7 ф Ħ R/S ₽gl == Ħ ຜ ტ ტ Ø Z Ŋ ტ

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9

## H6FXtripa fusion protein

1	M	G	S	Н	H	Н	H	H	H	G	S	I	Q	G	R	S	P	G	T	E	P	P	T	Q	K	P	K	K	I	V.	30
31	N	A	ĸ	K	D	V	v	N	T	K	M	F	E	E	L	K	s	R	L	D <sub>.</sub>	T	L	A	Q	E	v	A	L	L	ĸ	60
61	E	o	o	Α	L	0	т	v	S	·L	K	G	S	*																	73

# **H6FXtripB** fusion protein

1 M G S H H H H H H G S I Q G R S P G T E P P T Q K P K K I V 30
 31 N A K K D V V N T K M F E E L K S R L D T L A Q E V A L L K 60
 61 E Q Q A L Q T G S \*

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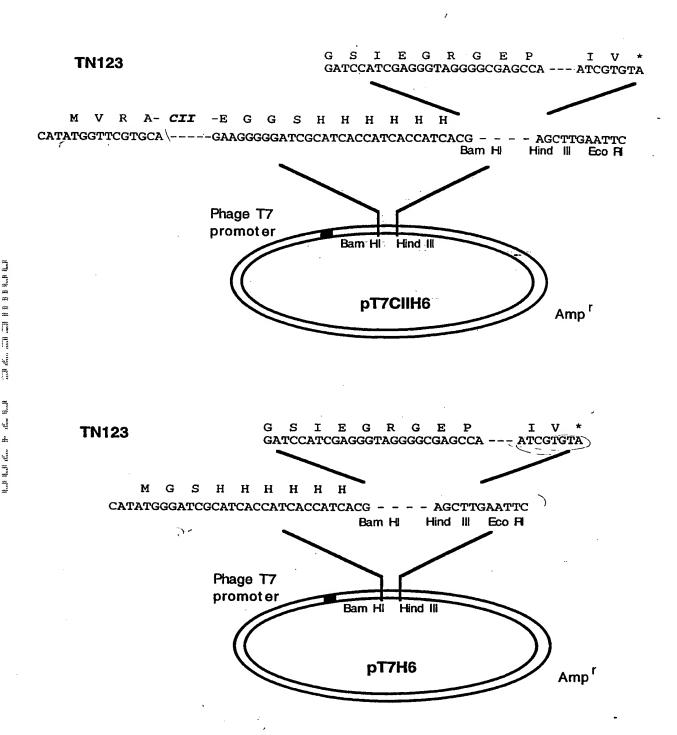


Fig. 5

#### CIIH6FXTN123 fusion protein

1 M V R A N K R N E A L R I E S A L L N K I A M L G T E K T A 30
31 E G G S H H H H H H G S I E G R G E P P T Q K P K K I V N A 60
61 K K D V V N T K M F E E L K S R L D T L A Q E V A L L K E Q 90
91 Q A L Q T V C L K G T K V H M K C F L A F T Q T K T F H E A 120
121 S E D C I S R G G T L S T P Q T G S E N D A L Y E Y L R Q S 150
151 V G N E A E I W L G L N D M A A E G T W V D M T G A R I A Y 180
181 K N W E T E I T A Q P D G G K T E N C A V L S G A A N G K W 210
228

## H6FXTN123 fusion protein

1 MGSHHHHHHGSIEGRGEPPTQKPKKIVNAK 30
31 KDVVNTKMFEELKSRLDTLAQEVALLKEQQ 60
61 ALQTVCLKGTKVHMKCFLAFTQTKTFHEAS 90
91 EDCISRGGTLSTPQTGSENDALYEYLRQSV 120
121 GNEAEIWLGLNDMAAEGTWVDMTGARIAYK 150
151 NWETEITAQPDGGKTENCAVLSGAANGKWF 180

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GSIEGRGEPP **TN12** GÁTCCATCGAGGGTAGGGGCGAGCCACCA ---- CAGACGGTCTA 1 7/ G S I Q G R V V N T GIV **TN23** GATCCATCCAGGGTAGGGTTGTGAACACA ---- GGGATCGTGTA 10 G S I E G R A TL Q GIV \* **TN3** GATCCATCGAGGGTAGGGCCCTGCAG ---- GGGATCGTGTA **зинини** CATATGGGATCGCATCACCATCACG - - - - AGCTTGAATTC Bam HI Hind III Eco FI Phage T7 promot er рТ7Н6 Amp <sup>r</sup>

Fig. 7

## **H6FXTN12** fusion protein

1	M	G.	S	H	H	H	H	H	H	G	S	Ι	E	G	R	G	E	P	P	T	Q	K	P	K	K	I	V	N	A	K	30
31	ĸ	D	v	v	N	т	K	M	F	E	E	L	ĸ	s	R	L	D	Т	L	A	Q	E	V	A	L	L	K	E	Q	Q	60
61	A	L	Q	т	v	*																									65

# H6FXTN23 fusion protein

1	M	G	S	Н	H	н	H	Н	H	G	s	I	Q	G	R	v	V	N	T	K	M	F	E	E	L	K	S	R	L	D	30
31	T	L	A	Q	E	v	A	L	L	ĸ	E	Q	Q	A	L	Q	Т	v	С	L	K	G	T	K	V	H	M	K	С	F	60
61	L	A	F	T	Q	T	ĸ	T	F	н	E	A	s	E	D	С	I	s	R	G	G	T	L	s	Т	P	Q	T	G	S	90
91	E	N	D	A	L	Y	E	Y	L	R	Q	s	v	G	N	E	A	E	I	W	L	G	L	N	D	M	A	A	E	G	120
.21	T	W	v	D	M	T	G	A	R	I	A	Y	K	N	W	E	T	E	I	T	A	Q	P	D	G	G	ĸ	T	E	N	150
.51	C	A	v	L	S	G	A	A	N	G	ĸ	W	F	D	ĸ	R	С	R	D	Q	L	P	Y	I	С	Q	F	G	I	V	180
.81	*																														

## **H6FXTN3** fusion protein

1	M	G	S	H	H	H	H	H	H	G	S	I	E	G	R	A	L	Q	T	V	C	L	K	G	Т	K	V	Н	M	K	30
31	С	F	L	A	F	T	Q	т	K	T	F	н	E	A	s	E	D	С	I	s	R	G	G	T	L	S	Т	P	Q	T	60
61	G	s	E	N	D	A	L	Y	E	Y	L	R	Q	s	v	G	N	E	A	E	I	W	L	G	L	N	D	M	A	A	90
91	E	G	T	W	v	D	M	T	G	A	R	I	A	Y	ĸ	N	W	E	T	E	I	Т	A	Q	P	D	G	G	K	Т	120
121	E	N	С	A	v	L	s	G	A	A	N	G	K	W	F	D	ĸ	R	С	R	D	Q	L	P	Y	I	С	Q	F	G	150
151	I	v	*																												152

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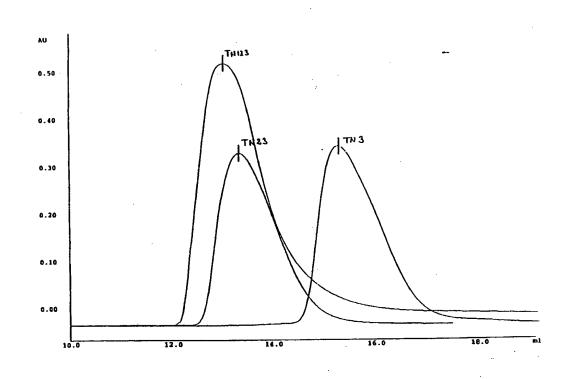


Fig. 9

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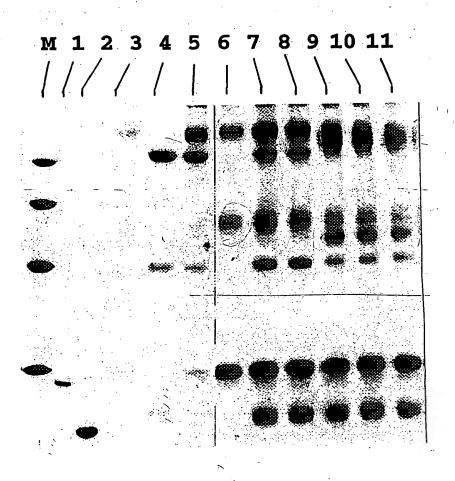


Fig. 10

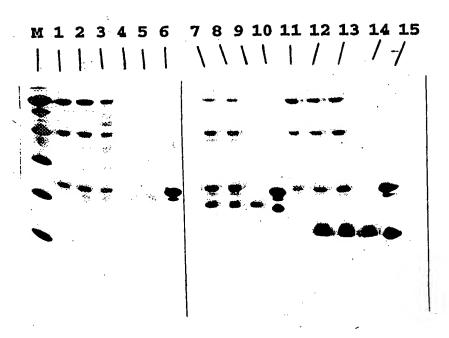


Fig. 11

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Fig. 12

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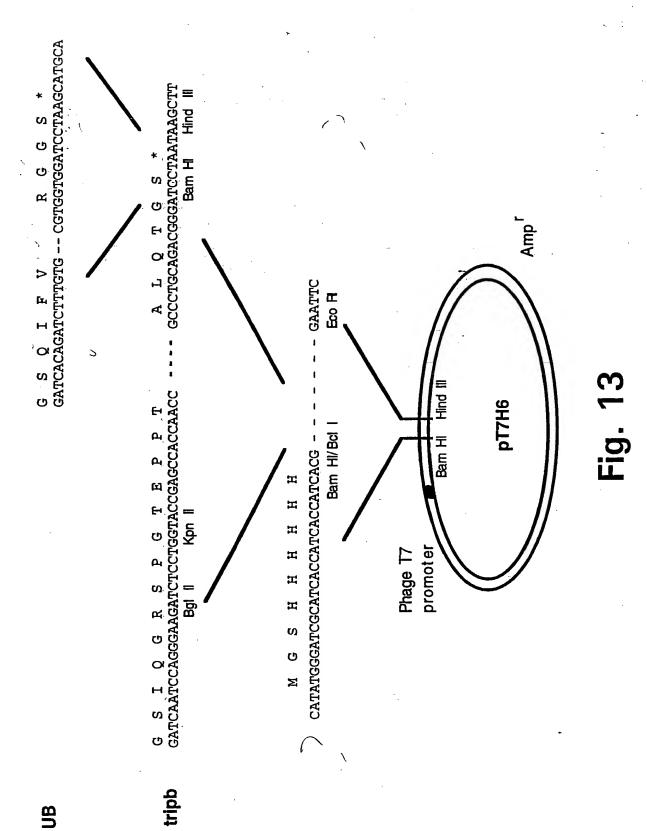


Fig. 13

### **H6FXtripb-UB** fusion protein

1 MGSHHHHHHGSIQGRSPGTEPPTQKPKKIV 30
31 NAKKDVVNTKMFEELKSRLDTLAQEVALLK 60
61 EQQALQTGSQIFVKTLTGKTITLEVEPSDT 90
91 IENVKAKIQDKEGIPPDQQRLIFAGKQLED 120
121 GRTLSDYNIQKESTLHLVLRLRGGS \* 145

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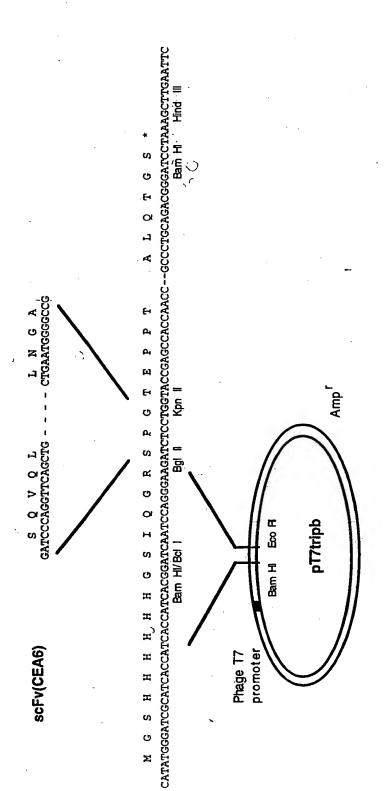
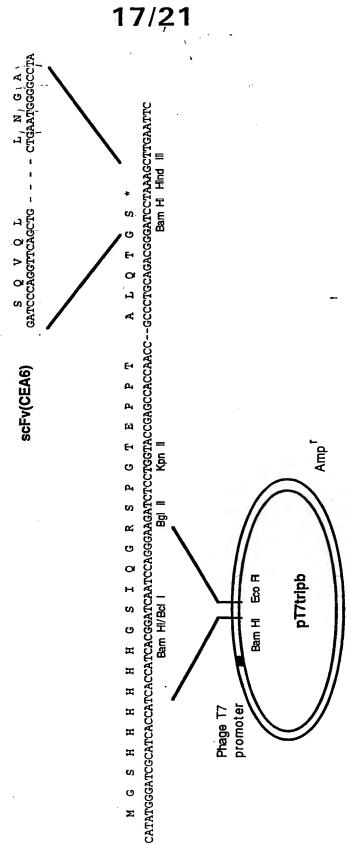


Fig. 15

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#### H6FXscFv(CEA6)-tripb fusion protein



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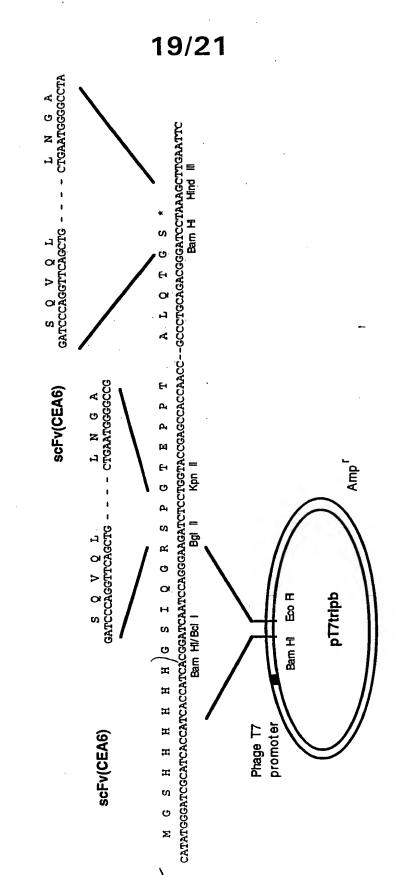
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## H6FXtripb-scFv(CEA6) fusion protein

1 M G S H H H H H H G S I Q G R S P G T E P P T Q K P K K I V 30 31 NAKKDVVNTKMFEELKSRLDTLAQEVALLK 60 61 E Q Q A L Q T G S Q V Q L Q Q S G A E V K K P G S S V K V S 90 91 C K A S G G T F S N S P I N W L R Q A P G Q G L E W M G S I 120 121 I P S F G T A N Y A O K F O G R L T I T A D E S T S T A Y M 150 151 E L S S L R S E D T A V Y Y C A G R S H N Y E L Y Y Y Y M D 180 181 V W G Q G T M V T V S S G G G G G G G G G G G G G D I Q 210 211 M T Q S P S T L S A S I G D R V T I T C R A S E G I Y H W L 240 241 AWYQQKPGKAPKLLIYKASSLASGAPSRFS 270 271 G S G S G T D F T L T I S S L Q P D D F A T Y Y C Q Q Y S N 300 301 Y P L T F G G G T K L E I K R A A A E Q K L I S E E D L N G 330 331 A \*





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#### H6FXscFv(CEA6)-tripb-scFv(CEA6) fusion protein

1 M G S H H H H H H G S I Q G R S Q V Q L Q Q S G A E V K K P 31 G S S V K V S C K A S G G T F S N S P I N W L R Q A P G Q G 61 LEWMGSIIPSFGTANYAQKFQGRLTITADE 90 91 S T S T A Y M E L S S L R S E D T A V Y Y C A G R S H N Y E 120 121 L Y Y Y Y M D V W G Q G T M V T V S S G G G G S G G G S G 150 151 G G G S D I Q M T Q S P S T L S A S I G D R V T I T C R A S 180 181 EGIYHWLAWYQQKPGKAPKLLIYKASSLAS 210 211 GAPSRFSGSGSGTDFTLTISSLQPDDFATY 240 241 Y C Q Q Y S N Y P L T F G G G T K L E I K R A A E Q K L I 270 271 SEEDLNGAGTEPPT'QKPKKIVNAKKDVVNT 300 301 KMFEELKSRLDTLAQEVALLKEQQALQTGS330 331 Q V Q L Q Q S G A E V K K P G S S V K V S C K A S G G T F S 360 361 NSPINWLRQAPGQGLEWMGSIIPSFGTANY 390 391 AQKFQGRLTITADESTSTAYMELSSLRSED 420 421 TAVYYCAGRSHNYELYYYYMDVWGQGTMVT 450 451 V S S G G G G G G G G G G G G G G D I Q M T Q S P S T L S 480 481 ASIGDRVTITCRASEGIYHWLAWYQQKPGK 510 511 APKLLIYKASSLASGAPSRFSGSGSGTDFT 540 541 LTISSLQPDDFATYYCQQYSNYPLTFGGGT 570 571 KLEIKRAAAEQKLISEEDLNGA \* 592

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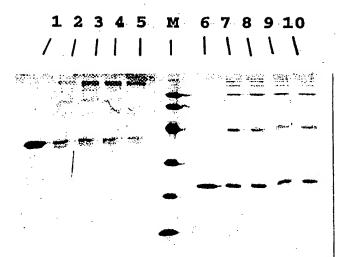


Fig. 21

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